

In the Claims:

Please cancel claims 2-36 and 42-49. Please add new claims 50-92.

Claim 1 (CURRENTLY AMENDED). A ligase-mediated method of recombination ~~creating at least one recombinant polynucleotide sequence~~, comprising:

- (a) providing oligonucleotide fragments derived from ~~an initial bank~~ each of at least two heterologous polynucleotide sequences of a polynucleotide bank;
- (b) hybridizing the fragments to an assembly matrix so that the hybridized fragments are oriented for ligation with each other; and
- (c) ligating the oriented hybridized fragments having immediately adjacent ends with a ligase to form a recombinant polynucleotide sequence.

Claims 2-36 (CANCELLED).

Claim 37. (WITHDRAWN) A recombinant polynucleotide sequence offering one or several advantageous characteristics compared to corresponding characteristics of reference sequences, obtained and selected by a process according to any one of claims 1 to 36, said sequence having a size greater than 1.5 Kpb.

Claim 38. (WITHDRAWN) A vector containing a polynucleotide sequence according to claim 37.

Claim 39. (WITHDRAWN) A cellular host transformed by a recombinant polynucleotide sequence according to claim 37 or by a vector according to claim 38.

Claim 40. (WITHDRAWN) A protein coded by a recombinant polynucleotide sequence according to claim 37.

Claim 41. (WITHDRAWN) A bank consisting of recombinant polynucleotide sequences according to claim 37, or of a vector according to claim 38, or of cellular hosts according to claim 39, or of proteins according to claim 40.

Claims 42-49 (CANCELLED).

Claim 50 (NEW). The method of claim 1, further comprising at least one repetition of the providing step, the hybridizing step or the ligating step.

Claim 51 (NEW). The method of claim 50, wherein the hybridizing step is repeated, before or after the ligating step, until at least a majority of the hybridized fragments have immediately adjacent ends.

Claim 52 (NEW). The method of claim 51, wherein, before a final ligating step, all of the hybridized fragments have immediately adjacent ends.

Claim 53 (NEW). The method of claim 1, wherein any polymerase extension performed during the hybridizing or ligating step, or between the hybridizing and ligating step, consists of gap filling between the hybridized fragments.

Claim 54 (NEW). The method of claim 1, wherein the method is performed without a polymerase.

Claim 55 (NEW). The method of claim 1, wherein the method is performed *in vitro*.

Claim 56 (NEW). The method of claim 1, wherein, at the providing step, the fragments are cleavage fragments.

Claim 57 (NEW). The method of claim 1, wherein, at the providing step, the fragments are random fragments.

Claim 58 (NEW). The method of claim 1, wherein the method of recombination is a method of random recombination.

Claim 59 (NEW). The method of claim 1, wherein the providing step comprises providing fragments that have been obtained in a controlled manner.

Claim 60 (NEW). The method of claim 1, wherein the providing step comprises fragmenting the at least two heterologous polynucleotide sequences in a controlled manner.

Claim 61 (NEW). The method of claim 1, wherein the at least two heterologous polynucleotide sequences differ from each other at more than one base position.

Claim 62 (NEW). The method of claim 61, wherein the at least two heterologous polynucleotide sequences are derived from at least two distinct genes.

Claim 63 (NEW) The method of claim 62, wherein the at least two heterologous polynucleotide sequences are derived from at least two distinct genes from at least two distinct gene families.

Claim 64 (NEW) The method of claim 62, wherein the at least two heterologous polynucleotide sequences are derived from at least two distinct genes from at least two different species of organism.

Claim 65 (NEW). The method of claim 1, wherein the at least two heterologous polynucleotide sequences are single-stranded.

Claim 66 (NEW). The method of claim 1, wherein at least one assembly matrix is double-stranded and it is first denatured and then added in single-stranded form at the hybridizing step.

Claim 67 (NEW). The method of claim 1, wherein at least one assembly matrix is single-stranded.

Claim 68 (NEW). The method of claim 1, wherein the ligase is a thermostable ligase that is active at high temperature.

Claim 69 (NEW). The method of claim 1, wherein the polynucleotide bank comprises artificial polynucleotide sequences.

Claim 70 (NEW). The method of claim 1, wherein, in addition to said fragments and assembly matrix, oligonucleotides of varying length, and single- or double-stranded, are added at the providing or hybridizing step.

Claim 71 (NEW). The method of claim 1, wherein the polynucleotide bank comprises a restricted bank.

Claim 72 (NEW). The method of claim 1, wherein the recombinant polynucleotide formed by the method is novel.

Claim 73 (NEW). The method of claim 1, further comprising cloning the recombinant polynucleotide sequence.

Claim 74 (NEW). The method of claim 1, wherein a fragment from the providing step is used as the assembly matrix.

Claim 75 (NEW). The method of claim 1, wherein the providing step comprises subjecting the at least two heterologous polynucleotide sequences to hydrolysis by the action of a plurality of different restriction enzymes or by the action of one or more restriction enzymes having a large number of cutting sites on the at least two heterologous polynucleotide sequences.

Claim 76 (NEW). The method of claim 75, wherein a fragment obtained at the providing step by a treatment with restriction enzymes is used as the assembly matrix.

Claim 77 (NEW). The method of claim 1, wherein the providing step further comprises randomly fragmenting the at least two heterologous polynucleotide sequences by treating them with DNAase I.

Claim 78 (NEW). The method of claim 77, wherein a fragment produced by the random fragmenting is used as the assembly matrix at the hybridizing step.

Claim 79 (NEW). The method of claim 1, wherein the hybridizing and ligating steps are performed simultaneously.

Claim 80 (NEW). The method of claim 1, wherein the at least two heterologous polynucleotide sequences are double-stranded and the providing step further comprises denaturing the fragments obtained at the providing step.

Claim 81 (NEW). The method of claim 1, further comprising selecting a recombinant polynucleotide sequence formed at the ligating step that exhibits advantageous characteristics compared to corresponding characteristics of one or more reference sequences.

Claim 82 (NEW). The method of claim 81, further comprising, after the selecting step, choosing at least one recombinant polynucleotide sequence formed at the ligating step and using the chosen sequence as a source of fragments or as an assembly matrix during at least one repetition of the providing or hybridizing step.

Claim 83 (NEW). The method of claim 81, further comprising separating the recombinant polynucleotide sequence formed at the ligating step from the assembly matrix.

Claim 84 (NEW). The method of claim 83, wherein the recombinant polynucleotide sequence is separated from the assembly matrix using a marker present on the assembly matrix or on the recombinant polynucleotide sequence.

Claim 85 (NEW). The method of claim 81, further comprising, before the selecting step, using polymerase extension to amplify the number of copies of the recombinant polynucleotide sequence.

Claim 86 (NEW). The method of claim 81, wherein the selected recombinant polynucleotide sequence is used to select a further recombinant polynucleotide sequence formed during a subsequent operation of the method.

Claim 87 (NEW). The method of claim 81, wherein the selection is performed by *in vitro* expression of the recombinant polynucleotide sequence.

Claim 88 (NEW). The method of claim 1, further comprising using a degrading enzyme at the hybridizing or ligating step that specifically recognizes and degrades any nonhybridized ends of the fragments when said nonhybridized ends overlap hybridized fragments on the assembly matrix.

Claim 89 (NEW). The method of claim 88, wherein the degrading enzyme is Flap endonuclease.

Claim 90 (NEW). The method of claim 88, wherein the degrading enzyme and the ligase are equally thermostable at the highest temperatures at which the steps of the method are conducted.

Claim 91 (NEW). The method of claim 88, wherein the degrading enzyme is a single-stranded exonuclease.

Claim 92 (NEW). A ligase-mediated method of recombination, comprising:

hybridizing oligonucleotide fragments derived from each of at least two heterologous polynucleotide sequences to an assembly matrix so that the hybridized fragments are oriented for ligation with each other; and

ligating the hybridized fragments having immediately adjacent ends with a ligase to form a recombinant polynucleotide sequence.